

approximately 800 nanoparticles/ $\mu\text{m}^2$ . Figure 39B: FE-SEM image of two layers of nanoparticles on the ITO slide. The average density of counted nanoparticles from 10 such images was approximately 2800 particles/ $\mu\text{m}^2$ . Figure 39C: Absorbance at 260 nm ( $A_{260}$ ) showing dissociation of a 0.5  $\mu\text{M}$  solution of the oligonucleotide duplex (**1** + **2** + **3**; see Figure 37, A) to single strands in 0.3 M NaCl, 10 mM phosphate buffer solution (pH 7). Figures 39D-F: Absorbance at 260 nm ( $A_{260}$ ) showing dissociation of 1 layer (Figure 39D), 4 layers (Figure 39E) and 10 layers (Figure 39F) of oligonucleotide-functionalized gold nanoparticles from glass slides immersed in 0.3 M NaCl, 10 mM phosphate buffer solution. Melting profiles were obtained by measuring the decreasing absorption at 520 nm ( $A_{520}$ ) through the slides with increasing temperature. In each of Figures 39D-F, the insets show the first derivatives of the measured dissociation curves. FWHM of these curves were (Figure 39C inset) 13.2  $^{\circ}\text{C}$ , (Figure 39D inset) 5.6  $^{\circ}\text{C}$ , (Figure 39E inset) 3.2  $^{\circ}\text{C}$ , and (Figure 39F inset) 2.9  $^{\circ}\text{C}$ .

**Figure 40:** Schematic diagram illustrating system used to measure the electrical properties of gold nanoparticle assemblies linked by DNA. For simplicity, only one hybridization event is drawn.

**Figure 41:** Schematic diagram illustrating a method of detecting nucleic acid using gold electrodes and gold nanoparticles.

## DETAILED DESCRIPTION OF THE PRESENTLY PREFERRED EMBODIMENTS

Nanoparticles useful in the practice of the invention include metal (*e.g.*, gold, silver, copper and platinum), semiconductor (*e.g.*, CdSe, CdS, and CdS or CdSe coated with ZnS) and magnetic (*e.g.*, ferromagnetite) colloidal materials. Other nanoparticles useful in the practice of the invention include ZnS, ZnO,  $\text{TiO}_2$ , AgI, AgBr,  $\text{HgI}_2$ , PbS, PbSe, ZnTe, CdTe,  $\text{In}_2\text{S}_3$ ,  $\text{In}_2\text{Se}_3$ ,  $\text{Cd}_3\text{P}_2$ ,  $\text{Cd}_3\text{As}_2$ , InAs, and GaAs. The size of the nanoparticles is preferably from about 5 nm to about 150 nm (mean diameter), more preferably from about 5 to about 50 nm, most preferably from about 10 to about 30 nm. The nanoparticles may also be rods.

Methods of making metal, semiconductor and magnetic nanoparticles are well-known in the art. See, e.g., Schmid, G. (ed.) *Clusters and Colloids* (VCH, Weinheim, 1994); Hayat, M.A. (ed.) *Colloidal Gold: Principles, Methods, and Applications* (Academic Press, San Diego, 1991); Massart, R., *IEEE Transactions On Magnetism*, **17**, 1247 (1981); Ahmadi, T.S. et al., *Science*, **272**, 1924 (1996); Henglein, A. et al., *J. Phys. Chem.*, **99**, 14129 (1995); Curtis, A.C., et al., *Angew. Chem. Int. Ed. Engl.*, **27**, 1530 (1988).

Methods of making ZnS, ZnO, TiO<sub>2</sub>, AgI, AgBr, HgI<sub>2</sub>, PbS, PbSe, ZnTe, CdTe, In<sub>2</sub>S<sub>3</sub>, In<sub>2</sub>Se<sub>3</sub>, Cd<sub>3</sub>P<sub>2</sub>, Cd<sub>3</sub>As<sub>2</sub>, InAs, and GaAs nanoparticles are also known in the art. See, e.g., Weller, *Angew. Chem. Int. Ed. Engl.*, **32**, 41 (1993); Henglein, *Top. Curr. Chem.*, **143**, 113 (1988); Henglein, *Chem. Rev.*, **89**, 1861 (1989); Brus, *Appl. Phys. A.*, **53**, 465 (1991); Bahncmann, in *Photochemical Conversion and Storage of Solar Energy* (eds. Pelizetti and Schiavello 1991), page 251; Wang and Herron, *J. Phys. Chem.*, **95**, 525 (1991); Olshavsky et al., *J. Am. Chem. Soc.*, **112**, 9438 (1990); Ushida et al., *J. Phys. Chem.*, **95**, 5382 (1992).

Suitable nanoparticles are also commercially available from, e.g., Ted Pella, Inc. (gold), Amersham Corporation (gold) and Nanoprobes, Inc. (gold).

Presently preferred for use in detecting nucleic acids are gold nanoparticles. Gold colloidal particles have high extinction coefficients for the bands that give rise to their beautiful colors. These intense colors change with particle size, concentration, interparticle distance, and extent of aggregation and shape (geometry) of the aggregates, making these materials particularly attractive for colorimetric assays. For instance, hybridization of oligonucleotides attached to gold nanoparticles with oligonucleotides and nucleic acids results in an immediate color change visible to the naked eye (see, e.g., the Examples).

Gold nanoparticles are also presently preferred for use in nanofabrication for the same reasons given above and because of their stability, ease of imaging by electron microscopy, and well-characterized modification with thiol functionalities (see below). Also preferred for use in nanofabrication are semiconductor nanoparticles because of their unique electronic and luminescent properties.

The nanoparticles, the oligonucleotides or both are functionalized in order to attach the oligonucleotides to the nanoparticles. Such methods are known in the art. For instance, oligonucleotides functionalized with alkanethiols at their 3'-termini or 5'-termini readily attach to gold nanoparticles. See Whitesides, *Proceedings of the Robert A. Welch Foundation 39th Conference On Chemical Research Nanophase Chemistry*, Houston, TX, pages 109-121 (1995). See also, Mucic et al. *Chem. Commun.* 555-557 (1996) (describes a method of attaching 3' thiol DNA to flat gold surfaces; this method can be used to attach oligonucleotides to nanoparticles). The alkanethiol method can also be used to attach oligonucleotides to other metal, semiconductor and magnetic colloids and to the other nanoparticles listed above. Other functional groups for attaching oligonucleotides to solid surfaces include phosphorothioate groups (see, e.g., U.S. Patent No. 5,472,881 for the binding of oligonucleotide-phosphorothioates to gold surfaces), substituted alkylsiloxanes (see, e.g. Burwell, *Chemical Technology*, 4, 370-377 (1974) and Matteucci and Caruthers, *J. Am. Chem. Soc.*, 103, 3185-3191 (1981) for binding of oligonucleotides to silica and glass surfaces, and Grabar et al., *Anal. Chem.*, 67, 735-743 for binding of aminoalkylsiloxanes and for similar binding of mercaptoalkylsiloxanes). Oligonucleotides terminated with a 5' thionucleoside or a 3' thionucleoside may also be used for attaching oligonucleotides to solid surfaces. The following references describe other methods which may be employed to attached oligonucleotides to nanoparticles: Nuzzo et al., *J. Am. Chem. Soc.*, 109, 2358 (1987) (disulfides on gold); Allara and Nuzzo, *Langmuir*, 1, 45 (1985) (carboxylic acids on aluminum); Allara and Tompkins, *J. Colloid Interface Sci.*, 49, 410-421 (1974) (carboxylic acids on copper); Iler, *The Chemistry Of Silica*, Chapter 6, (Wiley 1979) (carboxylic acids on silica); Timmons and Zisman, *J. Phys. Chem.*, 69, 984-990 (1965) (carboxylic acids on platinum); Soriaga and Hubbard, *J. Am. Chem. Soc.*, 104, 3937 (1982) (aromatic ring compounds on platinum); Hubbard, *Acc. Chem. Res.*, 13, 177 (1980) (sulfolanes, sulfoxides and other functionalized solvents on platinum); Hickman et al., *J. Am. Chem. Soc.*, 111, 7271 (1989) (isonitriles on platinum); Maoz and Sagiv, *Langmuir*, 3, 1045 (1987) (silanes on